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Reviewer: Durreshwar Anjum

Timestamp: [year=2008; month=3; day=10; hr=15; min=3; sec=9; ms=493; ]

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Application No: 10591751 Version No: 1.0

**Input Set:**

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**Total Warnings:** 2  
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**No. of SeqIDs Defined:** 18  
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<110> NAKAMURA, Hirofumi  
NAKANE, Akitaka  
KUBOTA, Hidetoshi

<120> B-FRUCTOFURANOSIDASE VARIANTS

<130> 2006\_1464A

<140> 10591751

<141> 2008-02-26

<150> JP 2004-060426

<151> 2004-03-04

<150> PCT/JP2005/003787

<151> 2005-03-04

<160> 18

<170> PatentIn version 3.3

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Leu Pro Asn Asn Thr Leu Phe His Val Trp Arg Pro Arg Ala His Ile  
20 25 30

ctg ccc gcc gag ggc cag atc ggc gac ccc tgc gcg cac tac acc gac 144  
Leu Pro Ala Glu Gly Gln Ile Gly Asp Pro Cys Ala His Tyr Thr Asp  
35 40 45

cca tcc acc ggc ctc ttc cac gtg ggg ttc ctg cac gac ggg gac ggc 192  
Pro Ser Thr Gly Leu Phe His Val Gly Phe Leu His Asp Gly Asp Gly  
50 55 60

atc gcg ggc gcc acc acg gcc aac ctg gcc acc tac acc gat acc tcc 240  
Ile Ala Gly Ala Thr Thr Ala Asn Leu Ala Thr Tyr Thr Asp Thr Ser  
65 70 75 80

gat aac ggg agc ttc ctg atc cag ccg ggc ggg aag aac gac ccc gtc 288  
Asp Asn Gly Ser Phe Leu Ile Gln Pro Gly Gly Lys Asn Asp Pro Val

| 85   | 90  | 95  |     |
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| gcc gtg ttc gac ggc gcc gtc atc ccc gtc ggc gtc aac aac acc ccc<br>Ala Val Phe Asp Gly Ala Val Ile Pro Val Gly Val Asn Asn Thr Pro |     |     | 336 |
| 100  | 105 | 110 |     |
| acc tta ctc tac acc tcc gtc tcc ttc ctg ccc atc cac tgg tcc atc<br>Thr Leu Leu Tyr Thr Ser Val Ser Phe Leu Pro Ile His Trp Ser Ile |     |     | 384 |
| 115  | 120 | 125 |     |
| ccc tac acc cgc ggc agc gag acg cag tcg ttg gcc gtc gcg cgc gac<br>Pro Tyr Thr Arg Gly Ser Glu Thr Gln Ser Leu Ala Val Ala Arg Asp |     |     | 432 |
| 130  | 135 | 140 |     |
| ggc ggc cgc cgc ttc gac aag ctc gac cag ggc ccc gtc atc gcc gac<br>Gly Gly Arg Arg Phe Asp Lys Leu Asp Gln Gly Pro Val Ile Ala Asp |     |     | 480 |
| 145  | 150 | 155 | 160 |
| cac ccc ttc gcc gtc gac gtc acc gcc ttc cgc gat ccg ttt gtc ttc<br>His Pro Phe Ala Val Asp Val Thr Ala Phe Arg Asp Pro Phe Val Phe |     |     | 528 |
| 165  | 170 | 175 |     |
| cgc agt gcc aag ttg gat gtg ctg ctg tcg ttg gat gag gag gtg gcg<br>Arg Ser Ala Lys Leu Asp Val Leu Leu Ser Leu Asp Glu Glu Val Ala |     |     | 576 |
| 180  | 185 | 190 |     |
| cgg aat gag acg gcc gtg cag cag gcc gtc gat ggc tgg acc gag aag<br>Arg Asn Glu Thr Ala Val Gln Gln Ala Val Asp Gly Trp Thr Glu Lys |     |     | 624 |
| 195  | 200 | 205 |     |
| aac gcc ccc tgg tat gtc gcg gtc tct ggc ggg gtg cac ggc gtc ggg<br>Asn Ala Pro Trp Tyr Val Ala Val Ser Gly Gly Val His Gly Val Gly |     |     | 672 |
| 210  | 215 | 220 |     |
| ccc gcg cag ttc ctc tac cgc cag aac ggc ggg aac gct tcc gag ttc<br>Pro Ala Gln Phe Leu Tyr Arg Gln Asn Gly Gly Asn Ala Ser Glu Phe |     |     | 720 |
| 225  | 230 | 235 | 240 |
| cag tac tgg gag tac ctc ggg gag tgg tgg cag gag gcg acc aac tcc<br>Gln Tyr Trp Glu Tyr Leu Gly Glu Trp Trp Gln Glu Ala Thr Asn Ser |     |     | 768 |
| 245  | 250 | 255 |     |
| agc tgg ggc gac gag ggc acc tgg gcc ggg cgc tgg ggg ttc aac ttc<br>Ser Trp Gly Asp Glu Gly Thr Trp Ala Gly Arg Trp Gly Phe Asn Phe |     |     | 816 |
| 260  | 265 | 270 |     |
| gag acg ggg aat gtg ctc ttc ctc acc gag gag ggc cat gac ccc cag<br>Glu Thr Gly Asn Val Leu Phe Leu Thr Glu Glu Gly His Asp Pro Gln |     |     | 864 |
| 275  | 280 | 285 |     |
| acg ggc gag gtg ttc gtc acc ctc ggc acg gag ggg tct ggc ctg cca<br>Thr Gly Glu Val Phe Val Thr Leu Gly Thr Glu Gly Ser Gly Leu Pro |     |     | 912 |
| 290  | 295 | 300 |     |
| atc gtg ccg cag gtc tcc agt atc cac gat atg ctg tgg gcg gcg ggt<br>Ile Val Pro Gln Val Ser Ser Ile His Asp Met Leu Trp Ala Ala Gly |     |     | 960 |
| 305  | 310 | 315 | 320 |

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|--|-----|-----|------|
| gag gtc ggg gtg ggc agt gag cag gag ggt gcc aag gtc gag ttc tcc  |     |     | 1008 |
| Glu Val Gly Val Gly Ser Glu Gln Glu Gly Ala Lys Val Glu Phe Ser  |     |     |      |
| 325  | 330 | 335 |      |
| ccc tcc atg gcc ggg ttt ctg gac tgg ggg ttc agc gcc tac gct gcg  |     |     | 1056 |
| Pro Ser Met Ala Gly Phe Leu Asp Trp Gly Phe Ser Ala Tyr Ala Ala  |     |     |      |
| 340  | 345 | 350 |      |
| gcg ggc aag gtg ctg ccg gcc agc tcg gcg gtg tcg aag acc agc ggc  |     |     | 1104 |
| Ala Gly Lys Val Leu Pro Ala Ser Ser Ala Val Ser Lys Thr Ser Gly  |     |     |      |
| 355  | 360 | 365 |      |
| gtg gag gtg gat cgg tat gtc tcg ttc gtc tgg ttg acg ggc gac cag  |     |     | 1152 |
| Val Glu Val Asp Arg Tyr Val Ser Phe Val Trp Leu Thr Gly Asp Gln  |     |     |      |
| 370  | 375 | 380 |      |
| tac gag cag gcg gac ggg ttc ccc acg gcc cag cag ggg tgg acg ggg  |     |     | 1200 |
| Tyr Glu Gln Ala Asp Gly Phe Pro Thr Ala Gln Gln Gly Trp Thr Gly  |     |     |      |
| 385  | 390 | 395 | 400  |
| tcg ctg ctg ctg ccg cgcc gag ctg aag gtg cag acg gtg gag aac gtc |     |     | 1248 |
| Ser Leu Leu Leu Pro Arg Glu Leu Lys Val Gln Thr Val Glu Asn Val  |     |     |      |
| 405  | 410 | 415 |      |
| gtc gac aac gag ctg gtg cgcc gag gag ggc gtg tcg tgg gtg gtg ggg |     |     | 1296 |
| Val Asp Asn Glu Leu Val Arg Glu Glu Gly Val Ser Trp Val Val Gly  |     |     |      |
| 420  | 425 | 430 |      |
| gag tcg gac aac cag acg gcc agg ctg cgcc acg ctg ggg atc acg atc |     |     | 1344 |
| Glu Ser Asp Asn Gln Thr Ala Arg Leu Arg Thr Leu Gly Ile Thr Ile  |     |     |      |
| 435  | 440 | 445 |      |
| gcc cggt gag acc aag gcg gcc ctg ctg gcc aac ggc tcg gtg acc gcg |     |     | 1392 |
| Ala Arg Glu Thr Lys Ala Ala Leu Leu Ala Asn Gly Ser Val Thr Ala  |     |     |      |
| 450  | 455 | 460 |      |
| gag gag gac cgcc acg ctg cag acg gcg gcc gtc gtg ccg ttc gcg caa |     |     | 1440 |
| Glu Glu Asp Arg Thr Leu Gln Thr Ala Ala Val Val Pro Phe Ala Gln  |     |     |      |
| 465  | 470 | 475 | 480  |
| tcg ccg agc tcc aag ttc ttc gtg ctg acg gcc cag ctg gag ttc ccc  |     |     | 1488 |
| Ser Pro Ser Ser Lys Phe Phe Val Leu Thr Ala Gln Leu Glu Phe Pro  |     |     |      |
| 485  | 490 | 495 |      |
| gcg agc gcg cgcc tcg tcc ccg ctc cag tcc ggg ttc gaa atc ctg gcg |     |     | 1536 |
| Ala Ser Ala Arg Ser Ser Pro Leu Gln Ser Gly Phe Glu Ile Leu Ala  |     |     |      |
| 500  | 505 | 510 |      |
| tcg gag ctg gag cgcc acg gcc atc tac tac cag ttc agc aac gag tcg |     |     | 1584 |
| Ser Glu Leu Glu Arg Thr Ala Ile Tyr Tyr Gln Phe Ser Asn Glu Ser  |     |     |      |
| 515  | 520 | 525 |      |
| ctg gtc gtc gac cgcc agc cag act agt gcg gcg gcg ccc acg aac ccc |     |     | 1632 |
| Leu Val Val Asp Arg Ser Gln Thr Ser Ala Ala Ala Pro Thr Asn Pro  |     |     |      |
| 530  | 535 | 540 |      |

|   |     |     |      |
|---|-----|-----|------|
| ggg ctg gat agc ttt act gag tcc ggc aag ttg cgg ttg ttc gac gtg |     |     | 1680 |
| Gly Leu Asp Ser Phe Thr Glu Ser Gly Lys Leu Arg Leu Phe Asp Val |     |     |      |
| 545   | 550 | 555 | 560  |
| atc gag aac ggc cag gag cag gtc gag acg ttg gat ctc act gtc gtc |     |     | 1728 |
| Ile Glu Asn Gly Gln Glu Gln Val Glu Thr Leu Asp Leu Thr Val Val |     |     |      |
| 565   | 570 | 575 |      |
| gtg gat aac gcg gtt gtc gag gtg tat gcc aac ggg cgc ttt gcg ttg |     |     | 1776 |
| Val Asp Asn Ala Val Val Glu Val Tyr Ala Asn Gly Arg Phe Ala Leu |     |     |      |
| 580   | 585 | 590 |      |
| agc acc tgg gcg aga tcg tgg tac gac aac tcc acc cag atc cgc ttc |     |     | 1824 |
| Ser Thr Trp Ala Arg Ser Trp Tyr Asp Asn Ser Thr Gln Ile Arg Phe |     |     |      |
| 595   | 600 | 605 |      |
| tcc cac aac ggc gag ggc gag gtg cag ttc agg aat gtc tcc gtg tcg |     |     | 1872 |
| Phe His Asn Gly Glu Gly Glu Val Gln Phe Arg Asn Val Ser Val Ser |     |     |      |
| 610   | 615 | 620 |      |
| gag ggg ctc tat aac gcc tgg ccg gag aga aat                     |     |     | 1905 |
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| Leu Pro Asn Asn Thr Leu Phe His Val Trp Arg Pro Arg Ala His Ile |     |     |      |
| 20  | 25  | 30  |      |
| Leu Pro Ala Glu Gly Gln Ile Gly Asp Pro Cys Ala His Tyr Thr Asp |     |     |      |
| 35  | 40  | 45  |      |
| Pro Ser Thr Gly Leu Phe His Val Gly Phe Leu His Asp Gly Asp Gly |     |     |      |
| 50  | 55  | 60  |      |
| Ile Ala Gly Ala Thr Thr Ala Asn Leu Ala Thr Tyr Thr Asp Thr Ser |     |     |      |
| 65  | 70  | 75  | 80   |
| Asp Asn Gly Ser Phe Leu Ile Gln Pro Gly Gly Lys Asn Asp Pro Val |     |     |      |
| 85  | 90  | 95  |      |
| Ala Val Phe Asp Gly Ala Val Ile Pro Val Gly Val Asn Asn Thr Pro |     |     |      |

|   | 100 | 105 | 110 |
|---|-----|-----|-----|
| Thr Leu Leu Tyr Thr Ser Val Ser Phe Leu Pro Ile His Trp Ser Ile |     |     |     |
| 115   | 120 | 125 |     |
| Pro Tyr Thr Arg Gly Ser Glu Thr Gln Ser Leu Ala Val Ala Arg Asp |     |     |     |
| 130   | 135 | 140 |     |
| Gly Gly Arg Arg Phe Asp Lys Leu Asp Gln Gly Pro Val Ile Ala Asp |     |     |     |
| 145   | 150 | 155 | 160 |
| His Pro Phe Ala Val Asp Val Thr Ala Phe Arg Asp Pro Phe Val Phe |     |     |     |
| 165   | 170 | 175 |     |
| Arg Ser Ala Lys Leu Asp Val Leu Leu Ser Leu Asp Glu Glu Val Ala |     |     |     |
| 180   | 185 | 190 |     |
| Arg Asn Glu Thr Ala Val Gln Gln Ala Val Asp Gly Trp Thr Glu Lys |     |     |     |
| 195   | 200 | 205 |     |
| Asn Ala Pro Trp Tyr Val Ala Val Ser Gly Gly Val His Gly Val Gly |     |     |     |
| 210   | 215 | 220 |     |
| Pro Ala Gln Phe Leu Tyr Arg Gln Asn Gly Gly Asn Ala Ser Glu Phe |     |     |     |
| 225   | 230 | 235 | 240 |
| Gln Tyr Trp Glu Tyr Leu Gly Glu Trp Trp Gln Glu Ala Thr Asn Ser |     |     |     |
| 245   | 250 | 255 |     |
| Ser Trp Gly Asp Glu Gly Thr Trp Ala Gly Arg Trp Gly Phe Asn Phe |     |     |     |
| 260   | 265 | 270 |     |
| Glu Thr Gly Asn Val Leu Phe Leu Thr Glu Glu Gly His Asp Pro Gln |     |     |     |
| 275   | 280 | 285 |     |
| Thr Gly Glu Val Phe Val Thr Leu Gly Thr Glu Gly Ser Gly Leu Pro |     |     |     |
| 290   | 295 | 300 |     |
| Ile Val Pro Gln Val Ser Ser Ile His Asp Met Leu Trp Ala Ala Gly |     |     |     |
| 305   | 310 | 315 | 320 |
| Glu Val Gly Val Gly Ser Glu Gln Glu Gly Ala Lys Val Glu Phe Ser |     |     |     |
| 325   | 330 | 335 |     |

Pro Ser Met Ala Gly Phe Leu Asp Trp Gly Phe Ser Ala Tyr Ala Ala  
340 345 350

Ala Gly Lys Val Leu Pro Ala Ser Ser Ala Val Ser Lys Thr Ser Gly  
355 360 365

Val Glu Val Asp Arg Tyr Val Ser Phe Val Trp Leu Thr Gly Asp Gln  
370 375 380

Tyr Glu Gln Ala Asp Gly Phe Pro Thr Ala Gln Gln Gly Trp Thr Gly  
385 390 395 400

Ser Leu Leu Leu Pro Arg Glu Leu Lys Val Gln Thr Val Glu Asn Val  
405 410 415

Val Asp Asn Glu Leu Val Arg Glu Glu Gly Val Ser Trp Val Val Gly  
420 425 430

Glu Ser Asp Asn Gln Thr Ala Arg Leu Arg Thr Leu Gly Ile Thr Ile  
435 440 445

Ala Arg Glu Thr Lys Ala Ala Leu Leu Ala Asn Gly Ser Val Thr Ala  
450 455 460

Glu Glu Asp Arg Thr Leu Gln Thr Ala Ala Val Val Pro Phe Ala Gln  
465 470 475 480

Ser Pro Ser Ser Lys Phe Phe Val Leu Thr Ala Gln Leu Glu Phe Pro  
485 490 495

Ala Ser Ala Arg Ser Ser Pro Leu Gln Ser Gly Phe Glu Ile Leu Ala  
500 505 510

Ser Glu Leu Glu Arg Thr Ala Ile Tyr Tyr Gln Phe Ser Asn Glu Ser  
515 520 525

Leu Val Val Asp Arg Ser Gln Thr Ser Ala Ala Ala Pro Thr Asn Pro  
530 535 540

Gly Leu Asp Ser Phe Thr Glu Ser Gly Lys Leu Arg Leu Phe Asp Val  
545 550 555 560

Ile Glu Asn Gly Gln Glu Gln Val Glu Thr Leu Asp Leu Thr Val Val

565

570

575

Val Asp Asn Ala Val Val Glu Val Tyr Ala Asn Gly Arg Phe Ala Leu

580

585

590

Ser Thr Trp Ala Arg Ser Trp Tyr Asp Asn Ser Thr Gln Ile Arg Phe

595

600

605

Phe His Asn Gly Glu Gly Glu Val Gln Phe Arg Asn Val Ser Val Ser

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15

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Ser Thr Leu Ala Asn Ala Ser Leu Phe Lys Thr Trp Arg Pro Arg Ala

20

25

30

cat ctt ctc cct cca tct ggg aac ata ggc gac ccg tgc ggg cac tat 144

His Leu Leu Pro Pro Ser Gly Asn Ile Gly Asp Pro Cys Gly His Tyr

35

40

45

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Thr Asp Pro Lys Thr Gly Leu Phe His Val Gly Trp Leu Tyr Ser Gly

50

55

60

att tcg gga gcg aca acc gac gat ctc gtt acc tat aaa gac ctc aat 240

Ile Ser Gly Ala Thr Thr Asp Asp Leu Val Thr Tyr Lys Asp Leu Asn

65

70

75

80

ccc gat gga gcc ccg tca att gtt gca gga gga aag aac gac cct ctt 288

Pro Asp Gly Ala Pro Ser Ile Val Ala Gly Gly Lys Asn Asp Pro Leu

85

90

95

tct gtc ttc gat ggc tcg gtc att cca agc ggt ata gac ggc atg cca 336

Ser Val Phe Asp Gly Ser Val Ile Pro Ser Gly Ile Asp Gly Met Pro  
100 105 110

act ctt ctg tat acc tct gta tca tac ctc cca atc cac tgg tcc atc 384  
Thr Leu Leu Tyr Thr Ser Val Ser Tyr Leu Pro Ile His Trp Ser Ile  
115 120 125

ccc tac acc cgg gga agc gag aca caa tcc ttg gcc gtt tcc tat gac 432  
Pro Tyr Thr Arg Gly Ser Glu Thr Gln Ser Leu Ala Val Ser Tyr Asp  
130 135 140

ggt ggt cac aac ttc acc aag ctc aac caa ggg ccc gtg atc cct acg 480  
Gly Gly His Asn Phe Thr Lys Leu Asn Gln Gly Pro Val Ile Pro Thr  
145 150 155 160

cct ccg ttt gct ctc aat gtc acc gct ttc cgt gac ccc tac gtt ttc 528  
Pro Pro Phe Ala Leu Asn Val Thr Ala Phe Arg Asp Pro Tyr Val Phe  
165 170 175

caa agc cca att ctg gac aaa tct gtc aat agt acc caa gga aca tgg 576  
Gln Ser Pro Ile Leu Asp Lys Ser Val Asn Ser Thr Gln Gly Thr Trp  
180 185 190

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